Light mediated activation of hypertrophy signaling in cardiac cells

Maya Groff, David Lawrence PhD, Lauren Haar PhD

Department of Chemical Biology and Medicinal Chemistry, Eshelman School of Pharmacy

THE UNIVERSITY of NORTH CAROLINA at CHAPEL HILL

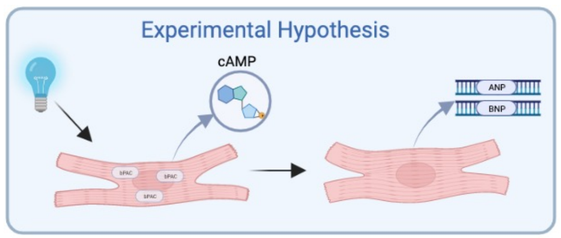
Introduction

Cyclic adenosine 3'5 monophosphate (cAMP) is a second messenger in cardiac growth signaling.[1]

cAMP induces cardiac hypertrophy but has also been linked to protection against heart failure.[2 3]

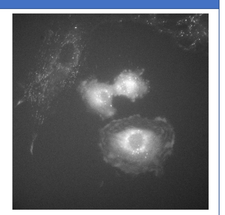
Optogenetic photoactivatable adenylyl cyclase from beggiatoa (bPAC) is a useful tool to induce cAMP with cellular and spatial-temporal control.[4]

Study Design



Procedure

Figure 1. bPAC with Cytosolic Expression. Widefield microscopic imaging (*60x oil, NA=1.35)* highlighting the cytosolic expression of bPAC transduced into H9c2 cells.



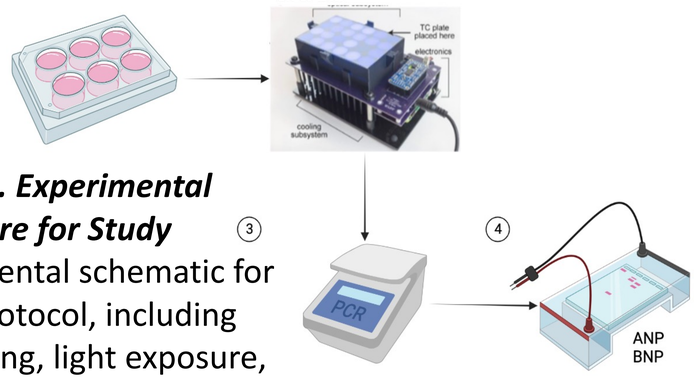


Figure 1. Experimental Procedure for Study Experimental schematic for study protocol, including cell plating, light exposure, polymerase chain reaction, and gel electrophoresis.

Results

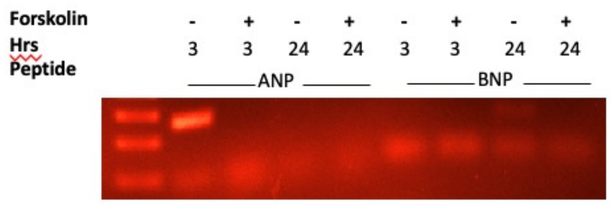


Figure 3. ANP and BNP production in forskolin treated bPAC cells. Gel electrophoresis of PCR product of bPAC cells treated with forskolin for 3 hrs (50 µM) and 24 hrs (25 µM).

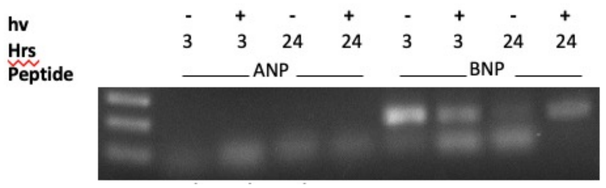


Figure 4. ANP and BNP production in light treated bPAC cells. Gel electrophoresis of PCR product of bPAC cells exposed to blue LED light for 1 s/hr for 3 hrs and 24 hrs.

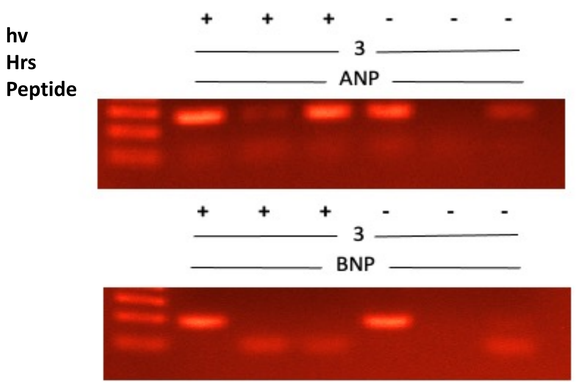
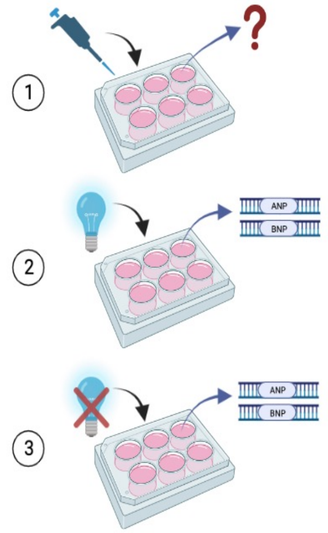


Figure 5. ANP and BNP production in light treated bPAC cells. Gel electrophoresis of PCR product for ANP (top) and BNP (bottom) of bPAC cells exposed to blue LED light for 1 s/hr for 3 hrs.

Conclusions



Further optimization of forskolin treatment conditions are needed (Fig 3).

ANP/BNP detected in light conditions, suggesting light-mediated cAMP activation (Fig 5).

ANP/BNP detected in dark conditions, suggesting dark activity (Fig 4).

Next Steps

Optimize forskolin treatment concentration for varying time points.

Optimal light-mediated cAMP activation conditions, exploring time and frequency of exposure.

Exploration of dark activity in bPAC.

References

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2. Zoccarato, A.; Surdo, N.; Aronsen, J.; et all. Cardiac Hypertrophy is Inhibited by a Local Pool of cAMP Regulated by Phosphodiesterase 2. *AHA Journals.* **2015.** 117 (8), 707- 719.
3. Zaccolo M. cAMP signal transduction in the heart: understanding spatial control for the development of novel therapeutic strategies. Br J Pharmacol. 2009 Sep;158(1):50-60. doi: 10.1111/j.1476-5381.2009.00185.x.
4. Naim N, Reece JM, Zhang X, Altschuler DL. Dual Activation of cAMP Production Through Photostimulation or Chemical Stimulation. Methods Mol Biol. 2020;2173:201-216. doi: 10.1007/978-1-0716-0755-8_14. PMID: 32651920; PMCID: PMC7968876.

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